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Reproduction of the cockle *Cerastoderma edule* (Linné, 1758) in the estuary of Oued Souss (southwestern Morocco)

Reproducción del berberecho *Cerastoderma edule* (Linné, 1758) en el estuario del Oued Souss (suroeste de Marruecos)

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ABSTRACT

Field and laboratory investigations were carried out from 2001 to 2003 on a population of *Cerastoderma edule* living in the estuary of Oued Souss, in order to determine the impact of domestic wastewater discharges on the biology of this species. During wastewater discharges, several episodes of partial spawning occurred from November to March, followed by another in April-May. The main spawning occurred nevertheless in August. The gonadic index was always higher than 1, so that the period of sexual rest was short. The period of recruitment (from June to October) was mainly correlated with spawning peaks. The renewal of the population was ensured by the recruitment of June-July, whereas young cockles recruited from August to October only represented 9.2% of the population at the end of autumn. Most 1-year-old cockles disappeared during the summer of the second year following their recruitment. After wastewater pollution stopped, an important recruitment of young cockles occurred. The spatial distribution of this species progressed upstream and the biomass of cockles had strongly increased. Because of its reproduction which takes place throughout the year, *C. edule,* as an opportunistic species, succeeded in colonizing this site.

RESUMEN

Se realizaron, entre 2001 y 2003, estudios de campo y de laboratorio sobre una población de *Cerastoderma edule* del estuario del Oued Souss, con el objetivo de determinar el impacto de los vertidos de aguas residuales sobre la biología de esta especie. Coincidiendo con los vertidos, se produjeron varios episodios de puesta parcial entre Noviembre y Marzo, seguidos de otro en Abril-Mayo. La puesta principal, sin embargo, se produjo en Agosto. El índice gonádico fue siempre superior a 1, indicando que el periodo de reposo sexual fue corto. El periodo de reclutamiento (de Junio a Octubre) estuvo esencialmente relacionado con picos de puesta. La reposición de la población fue asegurada por el reclutamiento de Junio-Julio, mientras berberechos jóvenes reclutados entre Agosto y Noviembre representaban tan solo el 9,2% de la población a finales de otoño. La mayor parte de los berberechos con un año de edad desaparecieron durante el verano del segundo año después de su reclutamiento. Al finalizar los vertidos se produjo un importante reclutamiento de berberechos jóvenes. La especie se extendió río arriba y la bio-

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masa de berberechos aumento fuertemente. Por su reproducción que se extiende sobre todo el año, *C. edule*, como especie oportunista, fue exitosa en la colonización de este lugar.

KEY WORDS: *Cerastoderma edule*, estuary, Oued Souss river, pollution, recruitment, reproductive cycle, wastewater discharges.
PALABRAS CLAVE: *Cerastoderma edule*, estuario, Oued Souss, contaminación, reclutamiento, ciclo reproductor,

INTRODUCTION

In marine environment, many biotic and abiotic factors may influence the life cycle of bivalves. If temperature, salinity, food supply, and tidal exposure are the most important causes known to modulate development in mytilids (Seed, 1975), other elements, such as intraspecific competition or environmental contaminants, can result in great variations in growth rate of bivalves (Seed and Suchanek, 1992). These factors also have an effect on the reproduction, settlement, recruitment, and production of these molluscs so that their variations directly affect the development of beds for each species of bivalve (Widdows and Donkin, 1992).

Contrary to numerous ecological reports on the effects of contaminants on different marine bivalves, the recovery of mollusc populations after the disappearance of pollution has been less investigated.

The implantation of a wastewater purification plant since November 2002 along the estuary of Oued Souss, at 4 km from the mouth of the estuary, had resulted in the fact that the fresh water still running in the river and the decanted wastewater were no longer discharged in the estuary but diverted to another coastal site: M'Zar, located at 3 km south. For this reason, the estuary, upstream to the purification plant, was only swept by sea tide at the present time.

As there existed a population of *Cerastoderma edule* living in the estuary of the Oued Souss river, it was interesting to determine the physiological state of these cockles during the pollution period by wastewater and the changes which have occurred after this contamination stopped. In view of these objectives, the following two questions arose: Had the discharge of wastewater before November 2002 caused repercussions on the reproductive cycle of *C. edule* and on the dynamics of this population? What consequences did the presence of sea water in the estuary after November 2002 have on the spatial distribution and biomass of *C. edule*? To answer the first question, monthly investigations from January 2001 to December 2002 were carried out in the habitat of *C. edule* i) to follow the different stages of sexual maturity in males and females, and ii) to specify the development of the different generations and cohorts of cockles. To tackle the second question, two surveys in 2002 and 2003 during summer months were performed in the estuary of Oued Souss river.

This study complements other reports made by our research team in the same ecosystem (Mouneyrac, Pellerin, Moukrim, Ait Alla, Durou and Viault, 2005; Ait Alla, Mouneyrac, Durou, Moukrim and Pellerin, 2005; Ait Alla, Gillet, Deutsch, Moukrim and Bergayou, 2005; Bergayou and Moukrim, 2003 and Gillet, Gorman, Tallec, Moukrim, Mouloud, Anajjar, Ait Alla, Bergayou and Kaaya, 2003).

MATERIALS AND METHODS

The estuary of Oued Souss is located on the Atlantic coast, in southwestern Morocco and is subjected to an arid climate. The mouth is swept by an intense marine hydrodynamism, responsible of the presence of great

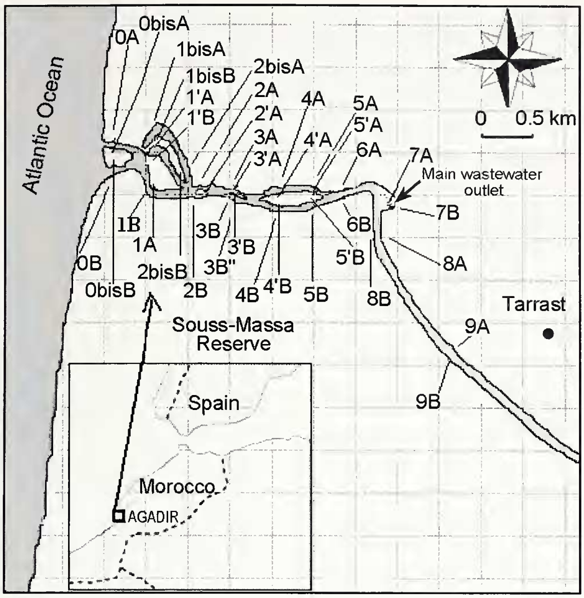

Figure 1. Location of sampling sites along the estuary of Oued Souss in the Bay of Agadir, southwestern Morocco.

Figura 1. Situación de las localidades de muestreo a lo largo del estuario del Oued Souss, en la bahía de Agadir, suroeste de Marruecos.

sandbanks in the estuary, with currents linked to high or low tides, and with high salinity (from 31.2 to 34.2‰).

Reproductive cycle

The study of reproductive cycle and population dynamics for *C. edule* were carried out at station 2 (30° 21.97′N north, 9° 35.98′W west), (Fig. 1). Selected because of its high biomass of the cockle population, this station was characterized by fine silty sand 2-5% of organic matter in the substratum and 340-440 mg/l of suspended matter in the water.

Two methods for studying the reproductive cycle of *C. edule* were used. The first was a classical histological study of gonads and was performed from January 2001 to March 2002. The second was a complementary study of condition index and microscopic examination of gonad smears (GUILLOU, BACHELET, AND GLÉMAREC, 1991) and was performed from August 2001 to August 2002. For the first and second study, respectively, 30 and 100 cockles (length, 20 to 30 mm) were collected by hand at low tide, at monthly intervals (respectively, a total of about 450 and 1300 accumulated cockles was examined).

For the first study (histological study of gonads), the shells of *C. edule* were opened and soft masses were prefixed in the Gendre's fixative for 24 h. In the laboratory, the shell of each bivalve was removed and small pieces of soft masses were post-fixed in a new solution of Gendre's fixative for 48 h before being dehydrated through a graded series of ethanol and butanol, and finally embedded in cytoparaffin (56°-58°C). Serial sections (thickness, 5 µm) were made before being stained with Gabe's trichrome, hemalun-eosin, or Mann-Dominici's method (GABE, 1968). The maturity of gonads was determined using the scale proposed by Lubet (1959) for *Mytilus edulis* (Table I).

The gonadic index (SEED, 1975) indicates the state of gonad maturity for

Table I. Terminology used by LUBET (1959) and LUCAS (1965) to study morphologically and histologically the gonadic developmental stages in bivalves.
Tabla I. Terminología empleada por LUBET (1959) y LUCAS (1965) para el estudio morfológico y histológico de estadios de desarrollo gonadal en bivalvos.

LUBET's scale (1959)	LUCAS's scale (1965)
Stage 0: sexual rest. Stage I: early gametogenesis with numerous goniae. Stage III D: spent, completely empty lumina.	Stage A: sex undetectable. This stage can correspond to sexual rest, previtellogenesis, or gamete resorption.
Stage II: actively developing gonads but mature gametes were not observed.	Stage B: sex detectable with difficulty to the naked eye.
Stage IIIA: near ripe follicles with mature gametes. Stage IIIB: spawning, follicles distended.	Stage C: identifiable gonad. The foot is salmon-stained in males and pearly white in females. Gonad maturation and spawning occurred during this stage.
Stage IIIC: partial spawning, partially empty lumina.	Stage D: gonadic reconstitution with co-existence of empty follicles and of tubules showing numerous goniae.

each population and is evaluated from histological slides. It was determined by giving a number to each of Lubet's gametogenic stages: stage 0 (number 1), stages I and II (2), stage IIIA (3), stages IIIB and IIIC (2), and stage IIID (1). For each sample of cockles, the number of gonads showing a gametogenic stage is multiplied by the corresponding number; the figures obtained were then added and the sum was then divided by the total number of cockles studied. This gonad index varied from 1 (all gonads were spent, with completely empty lumina) to 3 (all gonads were ripe).

The stereological analysis was made on the whole bivalves used for the histological examinations of gonads. For each *C. edule*, three histological slides were randomly chosen through the antero-posterior axis. The different cell categories present in gonadic follicles were counted on five ocular fields (magnification: x 100 for females, and x 400 for males) randomly selected in the visceral mass. In females, four categories: oogoniae, vitellogenic oocytes, ripe oocytes, and atresic oocytes, were considered. In males, the cells were classified into the following three categories: protogoniae and spermatogoniae, primary and secondary spermatocytes, and, lastly, spermatids and spermatozoa. The mean percentage of each cell category was calculated in relation to the total number of cells counted. Mean values and corresponding S.D. were established for each cell category.

For the second study, gonad smears and the determination of sex ratios were made by removing each bivalve from its shell and by rubbing soft masses against a histological slide. The microscopic examination was made using the scale of LUCAS (1965) adapted for *C. edule* by FERNANDEZ-CASTRO, GUILLOU, LE PENNEC, AND CARDENAS LOPEZ (1989) (see table I). The sex ratio (number of females in relation with the total number of bivalvs with a recognizable sex) was performed when this identification was easy. A χ^2 test was used to determine levels of statistical significance.

The condition index represents the variations of dry weight for a standard bivalve and aims to eliminate the effect of mollusc growth while revealing an accumulation or a loss of organic matter, associated with reproduction. A single size class (20-30 mm) was considered and 30 bivalves were randomly chosen to determine this condition index. The index selected was that proposed by LUCAS AND BENINGER (1985): IC = [(dry weight of soft masses) / (dry weight of valves) x 1000]. The dry weight was obtained using a dehydration of soft masses (or shell) in a desiccator (60°C, 24 h).

Population dynamics

To study the dynamics of the population and to specify the period of juvenile recruitment, samplings of *C. edule* were performed from January to December 2002. This study was also carried out in the station 2. Monthly, sixteen sediment samples (surface, 0.0625 m^2, height, 20 cm) were collected at low tide according to the method of quadrats used by ELLIOTT AND DECAMPS (1973, in BAYED, 1982). These samples were sieved (square meshes, 1 mm) to recover and count cockles. The antero-posterior length of each cockle was measured using a calliper rule (precision, 0.1 mm). For mollusc sizes less than 5 mm, a stereomicroscope equipped with a micrometric ocular was used. The individual values recorded for the length of bivalves and sampling dates were compared using the FISAT software (GAYANILO, SPARKE AND PAULY, 1996) to draw size histograms in relation to the frequency of animals and to make a modal analysis based on the algorithm according to the method by Battacharya (1967). This comparison, with the use of the χ^2 test (at $P<0.05$) allows to distinguish the different size classes in each monthly sample, to determine mollusc densities, and to specify recruitment periods.

Spatial distribution, density and biomass

To determine the spatial distribution, density, and biomass (ash-free dry weight, AFDW) of cockles, two surveys were carried out in 2002 and 2003 (during summer period). The replication of this study during these two periods (in 2002, when the estuary received the wastewater, and in 2003, when the discharge stopped) gives insights on the wastewater pollution impact and the reestablishment of the ecosystem when this pollution stopped. These investigations were made in a total of 8 stations (27 sampling sites) located at 400-500 m intervals from the estuary mouth up to the wastewater outlet (Fig. 1). In each site, four to six sediment samples (surface, 0.0625 m^2, height, 20 cm) were collected at low tide according to the method of quadrats used by ELLIOTT AND DECAMPS (1973, in BAYED, 1982). These samples were sieved (square meshes, 1 mm) to recuperate and count cockles, as above.

Ash-free dry weight (AFDW) was determined by dipping molluscs in a solution of 10% HCl until the shell was completely dissolved. Then, the soft parts were dried for 48 h at 80°C, weighed, placed at 600°C for 2 h and weighed again. The weight loss at 600°C was considered to represent the AFDW of individuals and was expressed in g/m^2 (BACHELET, BOUCHET AND LISSALDE, 1980-1981).

RESULTS

Effects of pollution on the biology of the species

Reproductive cycle: The study of gonad smears from August 2001 to August 2002 had allowed to recognize sex in 1138 bivalves (87.5%). Within this group, the sex ratio of *C. edule* was 51.1% ± 2.6%, and was well-balanced at 1:1 ($\chi^2 = 2.5$, $P < 0.05$). No hermaphrodite individuals were found in this population. In September, the A stage (undetectable sex) was observed in 75% of cockles and might correspond to a previtellogenic phase rather than a sexual rest, as the percentage of *C. edule* showing the C stage (gonad maturation) increased in October to reach about 50% in November. The frequency of C stage was above 50% between November and April (Fig. 2A), and subsequently decreased in May (from 75 to 20%), thus corresponding to a slight decrease of the

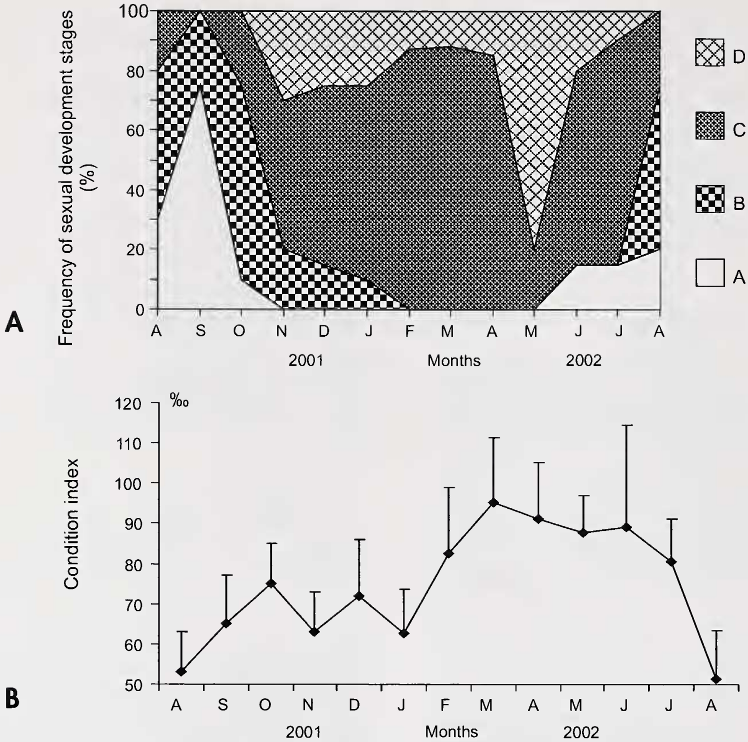

Figure 2. Distribution of sexual-development stages (A) according to the scale of LUCAS (1965) and annual cycle of condition indexes (B) in the populations of *C. edule* studied from August 2001 to August 2002. The cumulated frequencies of the different sexual-development stages corresponded to the total of bivalves studied (100%). For details of each stage, see Materials and Methods (Table I).

Figura 2. Distribución de los estadios de desarrollo sexual (A) según la escala de LUCAS (1965) y ciclo anual de índices de condición (B) en las poblaciones de C. edule *estudiadas entre agosto 2001 y agosto 2002. Las frecuencias acumuladas de distintos estadios de desarrollo sexual corresponden al total de bivalvos estudiados (100%). Véase Material y Métodos (Tabla I) para los detalles de cada estadio.*

condition index (Fig. 2B). An episode of partial spawning had thus occurred during this last period. In May, the frequency of the D stage (80%) proved that gametogenesis had resumed. In June and July, the C stage was the most frequent (65 to 75%) and was followed in August by a more marked decrease of the condition index (from 80 to 50‰) thus indicating an important release of gametes. It may be concluded that two periods of gamete maturation (C stage), the first ranging from November to April and the other occurring in June-July, were differentiated.

The histological study was carried out over a longer period of time (January 2001- March 2002) than the analysis of gonad smears. Mature gonads (IIIA stage) were often observed on more than 50% of cockles studied from January to April (Fig. 3). Episodes of partial spawning occurred from January to March; followed by another spawning in April and May (in May, the IIIB stage was noted in 60% of males, Fig. 3A). The reconstitution of the gonad (IIIC stage) is more marked in females (Fig. 3B) in May (80%) and seems more precocious in males. In both sexes, this stage developed in parallel from May to July so that partial spawning occurred during these months. In August, the IIIB stage was preponderant in both sexes (80% of

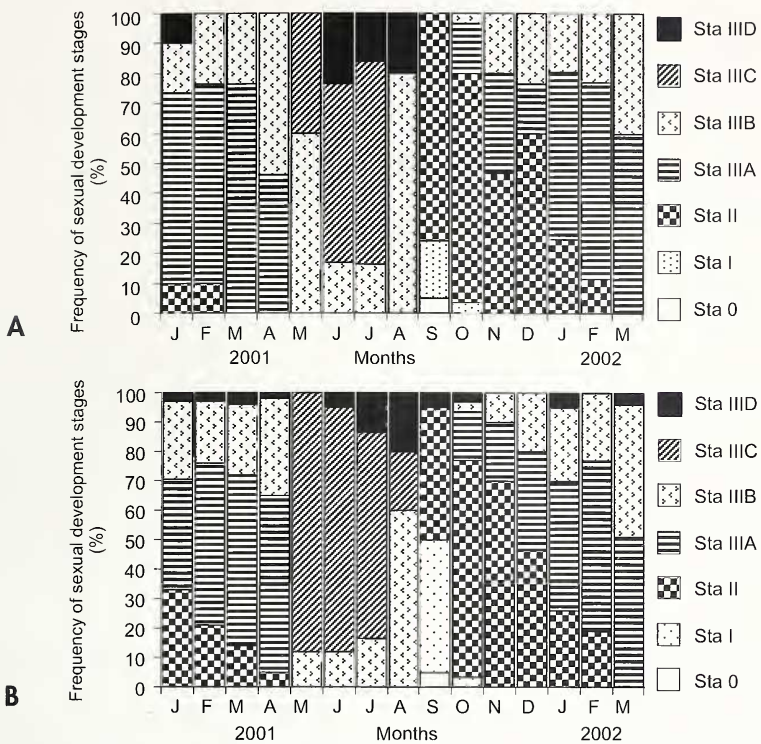

Figure 3. Distribution of gonadic-development stages over 2001-2002 in the males (A) and females (B) of *C. edule.* The cumulated frequencies of the different developmental stages corresponded to the whole bivalves studied (100%). For details of each stage, see Materials and Methods (Table I).
Figura 3. Distribución de los estadios de desarrollo gonadal sobre el periodo 2001-2001 en machos (A) y hembras (B) de C. edule. *Las frecuencias acumuladas de distintos estadios de desarrollo sexual corresponden al total de bivalvos estudiados (100%). Véase Material y Métodos (Tabla I) para los detalles de cada estadio.*

males, 60% of females), proving that the main spawning episode had happened. The resorption of gonads (IIID stage) was observed in a few individuals after the spawning of May and was less than 25% from June to August. Several females in IIID stage were noted during the other months, but they never exceeded 5%. In September, gametogenesis started in both sexes (I and II stages were found in 95% of cockles). The maturation of the gonad (IIIA stage) was observed in the first bivalves from October to December and partial spawning occurred from November to March.

The highest values (2.7) of the gonadic index (Fig. 4) were noted between January and April, thus confirming the presence of ripe gonads during this period. The lowest values (1.7) were found from June to August. In both sexes, the gonadic index was always higher than 1 so that the period of sexual rest was short.

Gametogenesis in the population of C. edule: Figure 5 gives the results of stereological analysis. From January to March, the percentages of mature gametes increased to peak in April. Another episode of gametogenesis began in May, as demonstrated by the high frequencies of young germinal cells during this month in both sexes. In males, the effect of this developing sper-

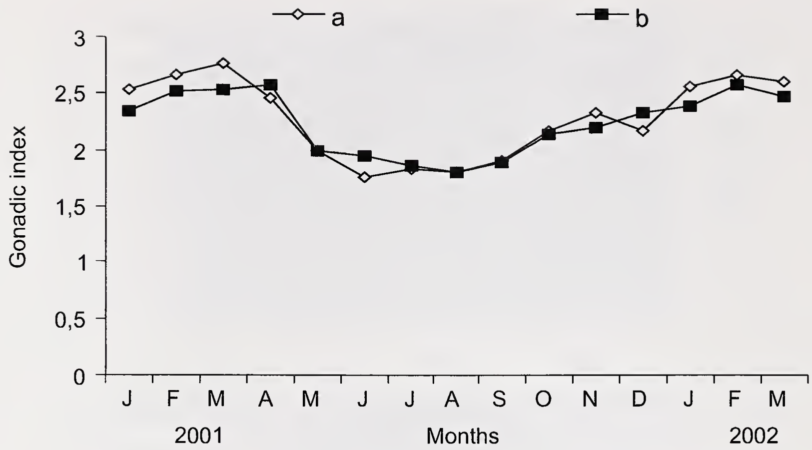

Figure 4. Annual cycle of the gonadic index in the males (A) and females (B) of *C. edule* over 2001-2002.
Figura 4. Ciclo anual del índice gonádico en machos (a) y hembras (b) de C. edule *en el periodo 2001-2002.*

matogenesis was high rates of spermatids and spermatozoa in June or August (grouped frequencies, 75% and 85%, respectively). In females, an important vitellogenesis was observed in May and June so that the highest frequency of mature oocytes (55%) was noted in July and that of degenerated oocytes (60%) in August. In September, the highest percentages of spermatogoniae (29%) in males and of oogoniae (37%) in females were noted, thus indicating the starting of another gametogenetic wave. From September to December, the gonadic tissue of males was important, as the grouped frequencies of spermatogoniae and of spermatocytes were more than 47%. The same finding was noted in females during this period (grouped rates of oogoniae and of vitellogenic oocytes, ≥70%). Low percentages of atresic oocytes (5-25%) were always observed in females throughout the year.

From the above study, it can be concluded that three successive gametogenetic waves were differentiated: the first one from January to April, the second one from May to July, and the third one from September to March. Ripe cockles for both sexes were predominant from January to April, in July, and from January to March.

Population dynamics: The lengths of *C. edule*, measured from January to December 2002, are given in Figure 6. The period of recruitment spanned from the end of spring to the onset of autumn. The first settlement of juvenile cockles occurred in June. A second cohort appeared in July, a third cohort in August, and a fourth one in October. At the end of autumn, the individuals recruited in June-July had a length of 18.3 ± 1.8 mm and constituted 71.6% of the population. By contrast, those originating from the grouped cohorts of August and October only had a length of 11.1 ± 1.1 mm and represented 9.2% of the population. In addition to these recruitments; a few young cockles (2-3%) were found for each date of sampling. In summer, there was a strong decrease in the number of cockles measuring 24.6 ± 1.9 mm in length. This diminution is followed by the almost complete disappearance of adult cockles just after the period of recruitment (August 2002) so that the population of *C. edule* in 2002 was mainly composed of young individuals which are being renewed each year.

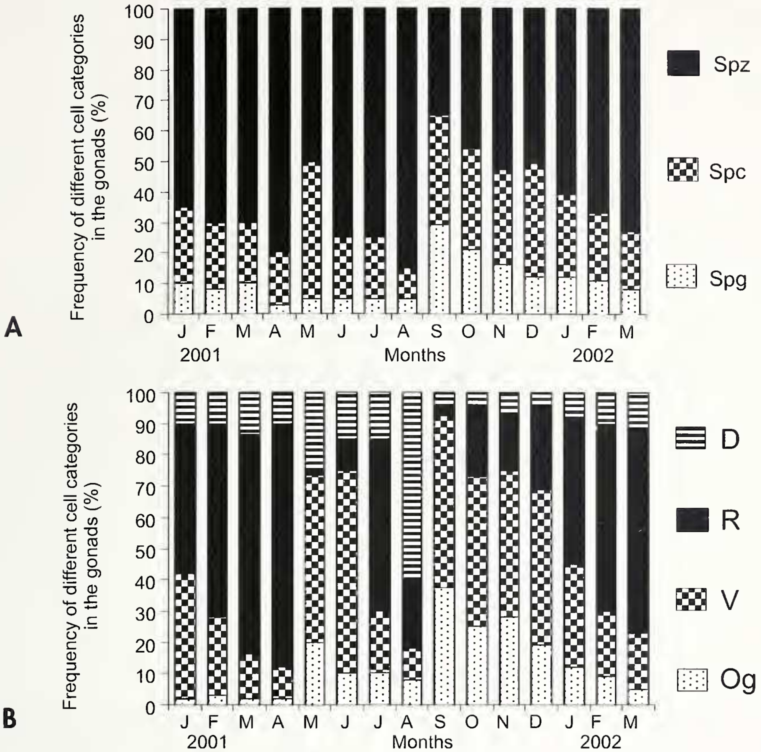

Figure 5. Frequencies of different cell categories in the male (A) and female (B) gonads of *C. edule* collected between January 2001 and March 2002. Male line: Spc (primary and secondary spermatocytes), Spg (protogoniae and spermatogoniae), Spz (spermatids and spermatozoa). Female line: D (degenerated oocytes), Og (oogoniae), R (mature oocytes), V (vitellogenic oocytes). The cumulated frequencies of the different cell categories corresponded to the whole bivalves studied (100%).
Figura 5. Frecuencia de distintas categorías de células en gónadas de machos (A) y hembras (B) de C. edule *recolectados entre enero 2001 y marzo 2002. En machos: Spc (Espermatocitos primarios y secundarios), Spg (protogonias y espermatogonias), Spz (espermatidios y espermatozoos); en hembras: D (ovocitos degenerados), Og (oogonias), R (ovocitos maduros), V (ovocitos vitelogénicos). Las frecuencias acumuladas de distintos estadios de desarrollo sexual corresponden al total de bivalvos estudiados (100%).*

Spatial distribution, biomass, and structure of population of C. edule before and after wastewater discharges

The spatial distribution of C. *edule* along the estuary, its density and its biomass are given in Table II for the summer surveys (2002 and 2003). During the period of wastewater discharge, the species was confined downstream (stations 1 and 2). By contrast, in 2003, its distribution had extended with an upstream penetration of cockles up to the station 5. The density of C. *edule* did not exceed 320 individuals/m^2 in 2002, and strongly increased in 2003 to reach values higher than 4,900 cockles/m^2 in several places (these values were found during the periods of recruitment). The mean biomass (AFDW) calculated on the cockle samples collected during summer surveys, increased from 2.54 g/m^2 in 2002 to 14.87 g/m^2 in 2003.

Table III shows the different groups of cockles constituting the population in summer surveys (2002 and 2003).

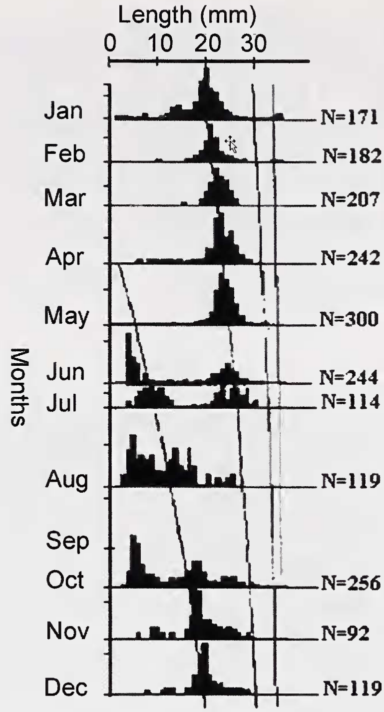

Figure 6. Size distribution of *C. edule* in the estuary of Oued Souss from January to December 2002 and the principal normal components. Month and numbers of individuals measured (N) are given for each sampling.

Figura 6. Distribución de tallas de C. edule *en el estuario del Oued Souss entre enero y diciembre 2002 y componentes normales principales. Meses y número de ejemplares medidos (N) están indicados para cada muestra.*

During the wastewater discharges in the estuary (July 2002), individuals are distributed among four groups: two classes with lowest lengths (6.81 ± 1.53 mm and 9.97 ± 0.83 mm), deriving from a recruitment in May and June; and two classes of old cockles (24,72 ± 2.17 mm and 27.5 ± 0,76 mm). In July 2003, the most frequent group (89.2%) was composed of young individuals (length, 6.55 ± 1.17 mm; 11.76 ± 1.63 mm) originating from a recruitment in May and June. Another group of medium size (36.25 ± 1.73 mm) appeared for the first time and probably comprised cockles more than one year old, as the absence of growth lines on cockle valves did not allow to easily identify the different age subgroups constituting this last size group.

DISCUSSION

The results reported in the present study on the gonochorism of *C. edule* agree with the reports of several authors (Gimazane, 1971; Kingston, 1974). Several cases of accidental hermaphrodism (4‰) were also noted by Fernandez Castro, Guillou, Le Pennec and Cardenas Lopez (1989). In the estuary of Oued Souss, the sex ratio of cockles was well-balanced so that pollution did not have an influence on the distribution of males and females. This finding agrees with studies that some authors have performed in other populations of *C. edule* at different latitudes, such as the report by Kingston (1974) along the coasts of Kent (UK), that of Ivell (1981)

Table II. Spatial distribution of *C. edule* along the estuary, its density and its Biomass (AFDWg /m²) during wastewater contamination (summer 2002) and after stopping of pollution (summer 2003).
Tabla II. Distribución espacial de C. edule *a lo largodel estuario, su densidad y su biomasa (AFDW/m²) durante vertidos de agua contaminada (verano 2002) y después de poner fin a la contaminación (verano 2003).*

Stations	Sampling sites	2002	2003	2002	2003
		Density (ind/m²)		Biomass AFDW (g/m²)	
0	0B		4		0.0012
	0bisA		872		32.368
	0bisB		24		0.08
1	1A	4		0.6492	
	1B		4		0.85
	1bis A		20		1.22
	1bisB	4	24	0.582	3.12
	1'A		36		0.446
	1'B		16		0.48
2	2A		108		0.28
	2B		6760		102.892
	2bisA	320		5.539	
	2bisB	112		3.648	
	2'A		4972		83.768
3	3A		8		0.1384
	3'A		432		3.7176
	3'B		636		6.8256
	3B		8		0.0416
4	4'A		16		0.074
	4'B		124		2.0676
5	5B		68		0.5248

in the Limfjord (Denmark), that of MEJUTO (1984) in Ria de Pasaxe (Spain), or still that of FERNANDEZ CASTRO, GUILLOU, LE PENNEC AND CARDENAS LOPEZ (1989) at Brouennou (France).

Through methods used for reproductive cycle analysis, complementary and concordant results were noted. In the year, two successive gametogenetic waves, the first occurring from September to April and the second in May-July, were differentiated in this population and spawning periods staggered over time. These phenomena can be explained by an effect of latitude (SOLA, 1997; RODRIGUEZ-RUA, PRADO, ROMEO AND BRUZON, 2003) and, in particular, by water temperature (HUGUES, 1971). When temperature was less than 10°C, it induced early spawning, followed by gonad reconstitution and a second period of spawning.

The presence of atresic oocytes observed throughout the year during vitellogenesis, as mentioned by LUBET (1991) is apparently a frequent phenomenon in bivalves noted in the start of gametogenesis (first mature oocytes degenerate), after partial spawnings and in the end of a reproductive cycle.

The long period of cockle recruitment (from June to October) in 2002 can

Table III. Distribution of shell lengths for C. edule during summer surveys (2002 and 2003): principal normal components.
Tabla III. Distribución de longitudes de conchas de C. edule *durante muestreos de verano (2002 y 2003): principales componentes normales.*

Group N⁰	Structure of population					
	July 2002			July 2003		
	Percentage (%)	Mean of shell length (mm)	S.D	Percentage (%)	Mean of shell length (mm)	S.D
1	19.5	6.81	1.53	11.4	6.55	1.17
2	16.4	9.97	0.83	77.8	11.76	1.63
3	55.5	24.72	2.17	10.68	36.25	1.73
4	8.4	27.5	0.76			

be easily explained by spawning which occurred from April to August and this time was perfectly consistent with the gonadic-development stages determined by the histological study of gonads and the examination of gonad smears. Contrary to juveniles recruited in June-July which had a better development, those settled from August to October showed a great mortality. This last result would not be related to wastewater pollution, in agreement with the reports by MADANI (1989), SAURIAU (1992), BACHELET, GUILLOU AND LABOURG (1992a), or with that by BACHELET, DESPREZ, DUCROTOY, GUILLOU, LABOURG, RYBARCZYK, SAURIAU, ELKAÏM AND GLÉMAREC (1992b). According to these authors, high mortalities of cockles were noted in the recruitments which occurred at the end of summer, in autumn, and in winter. This mortality affected small-sized (<10 mm) cockles and may be explained by the almost complete absence of energetic reserves (SAURIAU, 1992) whereas the metabolism of young cockles was changing (GABBOTT, 1976).

By contrast, the disappearance of medium-sized molluscs in 2002 coincided with the period of summer recruitment. This finding might be explained, either by a great predation of cockles by *Haematopus ostralegus* (this bird only tackled on medium sizes as reported by ATKINSON, CLARK, BELL, DARE, CLARK AND IRELAND, 2003; Johnstone and Norris, 2000), or by a high rate of mortality for these bivalves, as their vulnerability became more marked with increasing age and was also dependent on the quality of waters and/or an eventual eutrophication in the estuary, as demonstrated by DUCROTOY AND IBANEZ (2002). Indeed, as sea waters in winter and spring had high loads of mineral seston in the absence of pollution, the filter-feeders produced numerous pseudo-faeces and had a marked energetic expense for selective sorting of particles, mucus secretion, and cleaning of gills for *Mytilus edulis* (WIDDOWS, FIETH AND WORRALL, 1979) or for *Crassostrea gigas* (HÉRAL, 1986). According to FOSTER-SMITH (1975a, b) and NEWELL (1977), the behaviour of adult cockles was different, as they adapted their pumping activity in the presence of high concentrations of mineral seston, with a passage by a relative state of dormancy (SAVARI, LOCKWOOD AND SHEADER, 1991). However, such a behaviour did not seem to exist during wastewater discharges in summer, as cockles were in reproduction and had to live in waters double loaded with matters in suspension (pollution) and the phyto-planktonic bloom (with the increase of temperature), therefore placing these bivalves under stress conditions and inducing a high mortality.

Since November 2002, the discharge of decanted wastewater in another site beyond the estuary and the conversion of this ecosystem into a marine environment had resulted, for *C. edule*, in a wider spatial distribution, a strong increase of density, and the appearance of medium sizes in this site. To comment on these changes, it is necessary to take into account the reproduction of cockles living in this estuary, as it is stretched over all the year. Indeed, according to GORDO (1982), the reproductive cycle of Spanish and Portuguese populations of *C. edule* showed a period of sexual rest during summer months and the author explained it by the mean temperature of sea water which were more than 28°C and would inhibit the reproduction of this species, as they were close from the lethal temperature recorded for *C. edule* (ANSELL, BARNETT, BODOY AND MASSE, 1981). As the cockles from the estuary of the Oued Souss river practically did not have sexual rest and showed a gametogenesis starting towards autumn, the results noted in this site might be interpreted as an adaptation of this cockle population to high temperatures of sea waters which exist in the South of Morocco. Under these conditions, it is logical to consider *C. edule* an opportunistic species which might quickly overtake new areas in presence of favourable environmental conditions.

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